

Figure S1.

Cytotoxicity of recombinant ribosomal P-protein antigens. The viability of mouse spleen lymphocytes treated for 24-48h with different concentrations of P0 (A), P1 (B), P2 (C), P0-(P1-P2)₂ (D), and Msp-1₁₉ (E) was determined with the MTT assay. The results are expressed as the mean \pm S.E.M. of three independent experiments, each with eight separate cultures. *-significantly different from the 24h incubation time, $P \le 0.01$ (Wilcoxon's signed rank test).

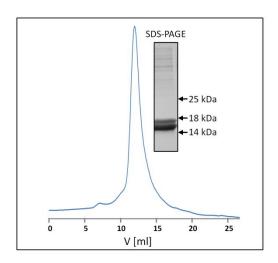


Figure S2.

Size-exclusion chromatography of the pentameric P-protein complex. Size-exclusion chromatography was performed using an analytical gel filtration column; the complex was eluted as a single symmetrical peak. Insert; SDS-PAGE analysis of protein fraction isolated

from size-exclusion chromatography.

Mice treated with	Th1 versus Tc		
_	r	Р	
P0	0.14	0.57	
P1	0.62	0.005	
P2	0.69	0.001	
P0-(P1-P2) ₂	0.92	0.0003	
Msp-1 ₁₉	0.91	0.0001	
adjuvant control mice	0.14	0.58	
untreated control mice	0.22	0.41	

Table S1.

Th1 lymphocytes support cellular immune response. The Spearman correlation between the number of the Th1 and Tc lymphocytes in the peripheral blood of mice immunized with the P0, P1, P2, P0-(P1-P2)₂, and MSP-1₁₉ proteins and in the adjuvant control mice as well as the untreated control mice. Each group consisted of eighteen mice. Values of $P \le 0.05$ were considered significant.

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	(CD4+CD25+ versus:				CD4+FoxP3+ versus:			
Protein	T	'c	Т	h1	Tc		Th1		
	r	P	r	P	r	P	r	Р	
P0	-0.046	0.85	0.13	0.59	0.17	0.49	0.2	0.42	
P1	-0.15	0.55	-0.13	0.56	0.31	0.2	0.3	0.22	
P2	-0.38	0.11	-0.82	0.00003	-0.63	0.004	-0.68	0.001	
P0-(P1-P2) ₂	-0.46	0.053	-0.49	0.039	-0.71	0.0008	-0.65	0.003	
Msp1 ₁₉	-0.43	0.07	054	0.02	-0.56	0.01	-0.48	0.04	
adjuvant control mice	0.28	0.24	-0.079	0.75	0.12	0.37	-0.1	0.62	
untreated control mice	0.3	0.11	0.1	0.66	0.26	0.21	0.15	0.54	

Table S2.

Regulation of cellular response (blood lymphocytes). The Spearman correlation between the numbers of CD4+CD25+ or CD4+FoxP3+ versus Tc and Th1 lymphocytes in the peripheral blood of mice immunized with the P0, P1, P2, P0-(P1-P2)₂, and Msp-1₁₉ proteins and in the adjuvant control mice as well as the untreated control mice. Correlations were calculated in each study group with eighteen mice. Values of $P \le 0.05$ were considered significant.

	С	CD4+FoxP3+ versus:					
Protein	T	'c	Th1				
	r	Р	r	P			
P0	0.45	0.22	0.31	0.4			
P1	-0.016	0.96	-0.26	0.48			
P2	-0.72	0.02	0.38	0.3			
P0-(P1-P2) ₂	-0.81	0.007	-0.85	0.003			
Msp-1 ₁₉	-0.76	0.017	-0.4	0.28			
adjuvant control mice	0.02	0.94	0.36	0.32			
untreated control mice	0.12	0.84	0.22	0.61			

Table S3.

Regulation of cellular response (spleen lymphocytes). The Spearman correlation between the numbers of CD4+FoxP3+ versus Tc and Th1 lymphocytes in cells isolated from the spleen of mice immunized with the P0, P1, P2, P0-(P1-P2)₂, and Msp-1₁₉ proteins and in the adjuvant control mice. Correlations were calculated in each study group with eighteen mice. Values of $P \le 0.05$ were considered significant.

	TGF-β versus:			IL-10 versus:					
Mice treated	CD4+CD25+ CD		CD4+l	CD4+FoxP3+ CI		CD4+CD25+		CD4+FoxP3+	
with	r	Р	r	P	r	P	r	P	
P0	-0.2	0.59	0.5	0.07	-	-	-	-	
P1	0.35	0.34	0.7	0.01	0.61	0.076	0.81	0.007	
P2	0.7	0.03	0.8	0.001	0.44	0.23	0.72	0.025	
P0-(P1-P2) ₂	0.82	0.005	0.51	0.01	0.66	0.049	0.5	0.16	
Msp-1 ₁₉	0.88	0.002	0.55	0.05	0.45	0.21	0.55	0.17	
adjuvant control mice	-0.15	0.49	0.1	0.61	0.11	0.33	0.2	0.21	
untreated control mice	-0.1	0.59	-0.19	0.69	0.17	0.84	0.11	0.66	

Table S4.

Interplay between regulatory blood lymphocytes and TGF- β or IL-10. The Spearman correlation between the numbers of serum TGF- β or IL-10 and CD4+CD25+ or CD4+FoxP3+ lymphocytes in the peripheral blood of mice immunized with the P0, P1, P2, P0-(P1-P2)₂, and Msp-1₁₉ proteins and in the adjuvant control mice. Correlations were calculated in each study group with eighteen mice. Values of $P \le 0.05$ were considered significant.

- the concentrations of circulating IL-10 levels in mice immunized with the P0 protein were below the detection threshold; therefore, calculation of Spearman correlations was not possible

Mice treated with	Th2 versus IL-10		
	r	P	
P0	-	-	
P1	0.49	0.18	
P2	0.65	0.05	
P0-(P1-P2) ₂	0.82	0.006	
Msp-1 ₁₉	0.81	0.007	
adjuvant control mice	0.15	0.54	
untreated control mice	0.21	0.41	

Table S5.

Production of IL-10 is associated with increased Th2 lymphocyte numbers. The Spearman correlation between the numbers of serum IL-10 and Th2 (CD4+CD30+) lymphocytes in the peripheral blood of mice immunized with the P0, P1, P2, P0-(P1-P2)₂, and Msp-1₁₉ proteins and in the adjuvant as well as the untreated control mice. Correlations were calculated in each study group with eighteen mice. Values of $P \le 0.05$ were considered significant.

- the concentrations of circulating IL-10 levels in mice immunized with the P0 protein were below the detection threshold; therefore, calculation of Spearman correlations was not possible

Mice treated with	total IgG versus Th2		
_	r	P	
P0	0.65	0.056	
P1	0.37	0.33	
P2	0.68	0.04	
P0-(P1-P2) ₂	0.91	0.0006	
Msp-1 ₁₉	0.89	0.001	
adjuvant control mice	0.21	0.26	
untreated control mice	0.2	0.33	

Table S6.

Humoral immune response support. The Spearman correlation between the IgG levels and the abundance of Th2 lymphocytes in the peripheral blood of mice immunized with the P0, P1, P2, P0-(P1-P2)₂, and Msp-1₁₉ proteins. For IgG determination, sera in each mouse group were pooled (n=9). Values of $P \le 0.05$ were considered significant.

Mice	IL-10 versus:					
immunized with	tota	l IgG	Th2			
With	r	r P		P		
P0	-	-	-	-		
P1	0.96	0.0004	0.49	0.18		
P2	0.95	0.0006	0.65	0.057		
P0-(P1-P2) ₂	0.69	0.037	0.82	0.006		
Msp-1 ₁₉	0.66	0.049	0.82	0.007		
adjuvant control mice	0.19	0.29	0.22	0.39		
untreated control mice	0.09	0.39	0.28	0.43		

Table S7.

IL-10 promotes humoral response. The Spearman correlation between IgG levels versus IL-10 serum concentrations and Th2 lymphocyte numbers in the peripheral blood of mice immunized with the P0, P1, P2, P0-(P1-P2)₂, and MSP-1₁₉ proteins and in the adjuvant control mice as well as the untreated control mice. For measurements of both IgG and IL-10, sera in each mice group were pooled (n=9). P0 did not induce detectable production of serum IL-10. Values of $P \le 0.05$ were considered significant.